# *Pathogen reduction requirements for direct potable reuse in Antarctica: evaluating human health risks in small communities*

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ABSTRACT

Small, remote communities often have limited access to energy and water. Direct potable reuse of treated wastewater has recently gained attention as a potential solution for water-stressed regions, but requires further evaluation specific to small communities. The required pathogen reductions needed for safe implementation of direct potable reuse of treated sewage is an important consideration but these are typically quantified for larger communities and cities. A quantitative microbial risk assessment (QMRA) was conducted, using norovirus, giardia and *Campylobacter* as reference pathogens, to determine the level of treatment required to meet the tolerable annual disease burden of $10^{-6}$ DALYs per person per year, using Davis Station in Antarctica as an example of a small remote community. Two scenarios were compared: published municipal sewage pathogen loads and estimated pathogen loads during a gastroenteritis outbreak. For the municipal sewage scenario, estimated required $\log_{10}$ reductions were 6.9, 8.0 and 7.4 for norovirus, giardia and *Campylobacter* respectively, while for the outbreak scenario the values were 12.1, 10.4 and 12.3 ($95^{th}$ percentiles). Pathogen concentrations are higher under outbreak conditions as a function of the relatively greater degree of contact between community members in a small population, compared with interactions in a large city, resulting in a higher proportion of the population being at risk of infection and illness. While the estimates of outbreak conditions may overestimate sewage concentration to some degree, the results suggest that additional treatment barriers would be required to achieve regulatory compliance for safe drinking water in small communities.

Keywords

*Campylobacter*, Drinking Water, Giardia, Norovirus, Quantitative Microbial Risk Assessment (QMRA), Sewage

Abbreviations

| | | |
|---|---|---|
| 47 | DALYs | disability adjusted life years |
| 48 | DPR | direct potable reuse |
| 49 | IPR | indirect potable reuse |
| 50 | LRV | $\log_{10}$ reduction values |
| 51 | QMRA | Quantitative Microbial Risk Assessment |

52

# 1. Introduction

Small remote communities sometimes struggle to adequately meet basic services such as power and water. In Australia, for example, there are many small remote communities. This is exemplified by the many remote indigenous communities, with nearly 13% of people living in the 838 communities with a population of less than 50 people and a significant number in communities with between 50 and 199 residents (ABS, 2008). More than half of the people living in remote indigenous communities rely on bore water as their main water source, 62% rely on community generators for electricity, only 30% are connected to a town sewerage system while 28% and 3.2% use septic tanks or pit toilets, respectively and high proportions of people experience interruptions in supply of services (ABS, 2008). In some of these communities, where water scarcity is an issue of concern, alternative sources of water may be needed. While recent droughts in Australia were accompanied by a drastic rise in the domestic use of grey water (ABS, 2007a; ABS, 2010a; ABS, 2010b), alternative sources of potable water have received less attention.

Indirect potable reuse schemes for the recycling of wastewater (IPR is the discharge of treated water into a receiving body prior to extraction and re-treatment for potable use) can be found in many countries; however, direct potable reuse (DPR is reuse without environmental mixing) is rare. There are currently only three DPR schemes in the world: Windhoek in Namibia (Lahnsteiner and Lempert, 2007), Cloudcroft in New Mexico and Big Springs in Texas (Tchobanoglous et al., 2011). While the more immediate driver of DPR is extreme water scarcity, various other factors also favour DPR systems, including whole-of-system life-cycle costs, reliability of water supply and quality and the exhaustion of economically feasible non-potable reuse options (Leverenz et al., 2011). An important consideration for system design and operation is the impact of population size on disease outbreaks, sewage quality and ultimately the required level of treatment. A greater understanding of these impacts is needed before the technology is implemented broadly.

79 Quantitative microbial risk assessment (QMRA) is a useful tool to assess pathogen reduction
80 requirements for wastewater recycling and has been used to inform the regulatory environment
81 relevant to wastewater schemes for non-potable reuse, IPR and DPR scenarios (NRMMC et al.,
82 2006b; NRMMC et al., 2008; NRMMC et al., 2009; WHO, 2006). Reuse guidelines are usually based
83 on water quality characteristics of municipal sewage from large cities and, using a tolerable annual
84 disease burden of $\leq 10^{-6}$ disability adjusted life years (DALYs) per person per year, QMRA has been
85 used to inform guidelines where recommended pathogen $\log_{10}$ reduction values (LRV) are presented
86 (NRMMC et al., 2008). Municipal sewage is typically of consistent or relatively stable quality, as a
87 function of the dilution effect from a large population base (NRMMC et al., 2008), although
88 differences between peak and non-peak seasons may be detectable; for example norovirus
89 concentrations in sewage may be up to 1 or 2 logs units higher during peak season (Katayama et al.,
90 2008; Nordgren et al., 2009; Victoria et al., 2010). Localized disease outbreaks and changes in
91 population size may significantly alter sewage microbial quality from a small population, potentially
92 affecting treatment requirements.
93
94 The objective of this study was to determine the required LRVs for DPR in small communities as this
95 has not been specifically considered in reuse guidelines. While any of a number of small remote
96 communities could have been chosen as a representative population for the model, Davis Station,
97 the largest of three permanent Australian research stations in Antarctica, was selected for this
98 exercise as there is current interest in DPR. The Australian Antarctic Division is undertaking a project
99 to reduce the environmental impact of sewage treatment and disposal at Davis Station. As part of
100 this project, research is being conducted into the potential implementation of DPR which, in addition
101 to providing a reliable potable water supply, could provide considerable energy savings as compared
102 with the existing water system. While Davis Station may not be a typical small community, only
103 minor modifications (volume of drinking water and days of exposure) would be required to

adequately reflect other populations. Regardless, the results of this assessment were considered generalizable to a range of other small communities, of which there are many in Australia and around the world.

# 2. Methods

The focus of this model was human health risks from waterborne pathogens, in particular diarrheal diseases, from ingestion of treated drinking water. Two complementary approaches were employed to estimate sewage pathogen concentrations: published values from municipal sewage treatment plants and estimated gastroenteritis outbreak conditions. Further detail is provided in supplementary materials.

## 2.1 QMRA

The QMRA method was used to determine required LRVs for direct potable reuse of wastewater starting from a health target—a tolerable annual burden of disease (*DB*) of $\leq 10^{-6}$ DALYs person$^{-1}$ year$^{-1}$—that has been widely adopted for both drinking water and non-potable reuse (NRMMC et al., 2006b; WHO, 2006; WHO, 2011). All model input parameters are listed in Table 1. Using the annual burden of disease calculation

$$DB = P_{\mathrm{ill}} B S_{\mathrm{f}}, \quad [1]$$

the tolerable annual probability of illness ($P_{\mathrm{ill}}$) was determined, where $B$ is the disease burden (DALYs per case of illness) and $S_{\mathrm{f}}$ is the proportion of the population susceptible to the disease.

While country-specific estimates of disease burden ($B$) are preferred, they are often non-existent. In this model, published values from a range of countries were used. For norovirus, a Uniform distribution (Cressey and Lake, 2009; Haagsma et al., 2008; Kemmeren et al., 2006; Lake et al., 2010; Masago et al., 2006) was used to represent the range of available values and similarly using Dutch data for giardia (Havelaar, 2012; Vijgen et al., 2007) and *Campylobacter* (Havelaar, 2012; Havelaar and Melse, 2003).

Disease susceptibility ($S_f$) is used to exclude the proportion of the population shown to be resistant to infection. There is evidence of resistance to norovirus infection (Johnson et al., 1990; Lindesmith et al., 2003; Teunis et al., 2008) related to both histo-blood group antigens and secretor status (Le Pendu et al., 2006) although it has been suggested that, due to the variation between norovirus genotypes, every person may be genetically susceptible to at least one norovirus genotype (Atmar, 2010). Since susceptibility to norovirus is uncertain, $S_f$ was represented by a Uniform distribution accounting for a range from secretor-positive individuals (0.8; Denborough and Downing, 1968; Thorven et al., 2005) through to all individuals (1.0). Despite many years of research, there remain many questions about the mechanisms of pathogenicity, host responses to infection and immunity to giardia infections (Roxström-Lindquist et al., 2006); therefore, in this work, all individuals were assumed susceptible ($S_f$ =1). No information on susceptibility to *Campylobacter* was found so the same assumption was made.

To estimate the tolerable daily probability of illness ($p_{ill}$), the original equation for annual probability of illness (WHO, 2006) was used such that

$$P_{\text{ill}} = 1 - (1 - p_{\text{ill}})^n, \quad [2]$$

for $n$ exposure events (days year$^{-1}$). In the model, the summer period (months where population >30) was assumed to be the period of exposure (due to the movement of people to and from the station) and was represented by a Uniform distribution determined from Davis Station records between 2005 and 2010. The tolerable daily probability of infection ($p_{inf}$) was determined using published dose-response models for norovirus (Teunis et al., 2008), giardia (Teunis et al., 1996) and campylobacter (Teunis et al., 2005). Full details of dose-response models and determination of tolerable dose are provided in supplementary materials.

152 Table 1. Model input parameters.

| Parameter | Units | Distribution or Point Estimates[a], [mean [b]] | References and Justification |
|---|---|---|---|
| Disease Burden (*B*) | DALYs case of illness$^{-1}$ | | |
| Norovirus | | Uniform($3.71\times10^{-4}$, $6.23\times10^{-3}$), [$3.30\times10^{-3}$] | (Cressey and Lake, 2009; Haagsma et al., 2008; Kemmeren et al., 2006; Lake et al., 2010; Masago et al., 2006) |
| Giardia | | Uniform($2.10\times10^{-3}$, $2.68\times10^{-3}$), [$2.39\times10^{-3}$] | (Havelaar, 2012; Vijgen et al., 2007) |
| *Campylobacter* | | Uniform($4.60\times10^{-3}$, $4.10\times10^{-2}$), [$2.28\times10^{-2}$] | (Havelaar, 2012; Havelaar and Melse, 2003) |
| Susceptibility fraction ($S_f$) | proportion | | |
| Norovirus | | Uniform(0.8, 1.0), [0.9] | (Atmar, 2010; Denborough and Downing, 1968; Soller et al., 2010; Thorven et al., 2005) |
| Giardia, *Campylobacter* | | 1 | |
| Exposure events (*n*) | days year$^{-1}$ | Uniform(62, 121), [91.5] | Total number of days for months with population ≥30 (AAD, 2011) between 2005 and 2010 |
| Dose-response models | | | |
| Norovirus (a+b inoculum) | | full Beta-Poisson: $\alpha_{NV}$=0.04, $\beta_{NV}$=0.055, $\eta_{NV}$=0.00255, *r*_NV=0.086, $a_{NV}$=0.9997 | (Teunis et al., 2008) |
| Giardia | | exponential: *r*_G=Triangular(0.0044, 0.0566, | (Teunis et al., 1996); min/max are 95$^{th}$ confidence intervals |

| | | | |
|---|---|---|---|
| | | 0.0199), [0.027] | |
| *Campylobacter* | | full Beta-Poisson: $\alpha$=0.024, $\beta$=0.011, $\eta_C$=3.63x10$^{-9}$, $r$_C=2.44x10$^{8}$ | (Teunis et al., 2005) |
| Giardia infection:illness (*inf:ill*) | proportion | Uniform(0.24, 0.93), [0.58] | (Birkhead and Vogt, 1989; Hoque et al., 2002; Lopez et al., 1980; Yakoob et al., 2010) |
| Daily water consumption (*V*) | L person$^{-1}$ | Lognormal(3, 1) – truncated at 2 and 6; $\mu$=1.05, $\delta$=0.32 | (Hunter et al., 2011; Roche et al., 2012; Schijven et al., 2011; USEPA, 2004; USEPA, 2006) |
| Sewage concentration – municipal sewage ($c_{sewage}$) | | | |
| Norovirus | PCR units L$^{-1}$ | Mixture (A, B), [3.12x10$^{6}$] | 11.1% recovery efficiency (Katayama et al., 2008) applied to A & B |
| | | A = Lognormal(2.19x10$^{6}$, 2.60x10$^{6}$); $\mu$=14.2, $\delta$=0.94 | (Katayama et al., 2008) |
| | | B = Lognormal(4.06x10$^{6}$, 6.27x10$^{6}$); $\mu$=14.6, $\delta$=1.11 | (Haramoto et al., 2006) |
| Giardia | cysts L$^{-1}$ | Mixture (G1, G2, G3), [2.51x10$^{3}$] | (Van Den Akker et al., 2011) |
| | | G1 = 10^Normal(2.90, 0.56) | recovery included in values (32-47%) |
| | | G2 = 10^Normal(2.94, 0.77) | |
| | | G3 = 10^Normal(2.57, 0.72) | |
| *Campylobacter* | cfu L$^{-1}$ | Lognormal(1.90x10$^{3}$, 5.00x10$^{3}$); $\mu$=6.51, $\delta$=1.44 | (NRMMC et al., 2006a) |

| | | | |
|---|---|---|---|
| Station population ($P$) | # people | Discrete distribution (min=51, max=106), [72] | daily station population in months with population ≥30; data from 2005-2011, n=601 (AAD, 2011) |
| Secondary attack rate ($A_r$) | proportion | | |
| Norovirus | | Uniform(0.14, 0.22), [0.18] | (Alfano-Sobsey et al., 2012; Baron et al., 1982; Götz et al., 2002; Johansson et al., 2002; ter Waarbeek et al., 2010) |
| Giardia | | Uniform(0.17, 0.18), [0.175] | (Katz et al., 2006; Pickering et al., 1981) |
| *Campylobacter* | | Uniform(0, 0.15), [0.075] | (Evans, 1996; Norkrans and Svedhem, 1982; Porter and Reid, 1980) |
| Peak shedding rate | | | |
| Norovirus ($S_{NV}$) | copies g-faeces$^{-1}$ | Uniform($2.9\times10^{10}$, $1.6\times10^{12}$), [$8.2\times10^{11}$] | (Atmar et al., 2008; Chan et al., 2006; Lee et al., 2007) |
| Giardia ($S_G$) | cysts person$^{-1}$ day$^{-1}$ | Uniform($6.42\times10^{8}$, $7.05\times10^{8}$), [$6.73\times10^{8}$] | (Tsuchiya, 1931) |
| *Campylobacter* ($S_C$) | cfu g-faeces$^{-1}$ | Uniform($10^4$, $10^9$), [$5\times10^8$] | (Feachem et al., 1983; Lin et al., 2008) |
| Daily diarrhoeal faecal weight ($F$) | g-faeces person$^{-1}$ | Uniform(200, 750), [475] | (Rao, 2006) |
| Daily water use ($W$) | L person$^{-1}$ day$^{-1}$ | Uniform(90, 174), [132] | Davis Station between 2010 and 2011 (AAD, 2011; AAD, 2012) |

[a]Distributions: Lognormal(mean, sd), values from 1,000,000 iterations, population parameters $\mu$ and $\delta$ calculated as follows: $\mu = \ln(\bar{x}) - 0.5\ln(1+s^2/\bar{x}^2)$, $\delta = [\ln(1+(s^2/\bar{x}^2))]^{1/2}$, where $\bar{x}$ is the sample mean and $s^2$ the sample standard deviation; Mixture is a set of random values drawn from each distribution with equal weighting; Normal(mean, sd); Triangular(min, max, mode/most likely); Uniform(min, max).

[b]mean of 1,000,000 iterations (for information purposes only).

158 The tolerable pathogen concentration in treated drinking water ($c_{\text{tolerable}}$; organisms $L^{-1}$) was
159 estimated from the exposure model,

$$\lambda = c_{tolerable}V, \quad [3]$$

160 using the estimated tolerable dose ($\lambda$) and the daily per capita water consumption ($V$; L $person^{-1}$
161 $day^{-1}$). Per capita water consumption at Davis Station is much higher than that of the general
162 population (typically assumed to be 2 L $day^{-1}$) as humidity is very low in Antarctica. Some community
163 members have indicated they drink much more than the recommended 4 L, with consumption of up
164 to 6 L per day considered quite reasonable. Variability in drinking water consumption was
165 represented using a Lognormal distribution (Ãstrom et al., 2007; Pintar et al., 2012; Schijven et al.,
166 2011) with a mean daily drinking water consumption of 3 L. In studies with mean daily drinking
167 water consumption greater than 1 L (Table S.1), standard deviations ranged from 0.8 to 1.2;
168 therefore, the middle value (1.0) was chosen to represent variation and the distribution was
169 truncated at the likely minimum and maximum values (2 and 6 L).
170
171 Finally, the required $\log_{10}$ reduction value (LRV) in sewage, necessary to meet tolerable drinking
172 water quality, was calculated as

$$\text{LRV} = log_{10}(c) - log_{10}(c_{\text{tolerable}}), \quad [4]$$

173 where the pathogen concentrations in sewage ($c$) were estimated using two different methods: 1)
174 published values of pathogen concentrations in municipal wastewater and 2) estimates of sewage
175 pathogen concentrations during a gastroenteritis outbreak at Davis Station. There was no available
176 information on concentrations of pathogens or indicator organisms in raw sewage at Davis Station.
177
178 *Norovirus,* giardia and *Campylobacter* concentrations in municipal wastewater ($c_{\text{sewage}}$; # $L^{-1}$) were
179 assumed to follow a Lognormal distribution, with values drawn from published literature (refer to
180 supplementary materials). An estimate of outbreak conditions at Davis Station was developed, with

an outbreak defined as the arrival of one infected person. Outbreak sewage pathogen concentrations ($c_o$; # $L^{-1}$) were estimated using the following equations

$$c_o = \frac{(1+PA_r)S}{WP}, \quad [5]$$

$$S = S_{NV}F \text{ or } S = S_C F, \quad [6]$$

where $P$ is the population on a given summer day, $A_r$ is the secondary attack rate (proportion), $S$ is the peak daily pathogen shedding rate (# $person^{-1}$ $day^{-1}$), $W$ is the per capita water use (L $person^{-1}$ $day^{-1}$), $S_{NV}$ and $S_C$ are the norovirus and *Campylobacter* shedding concentrations (# g $faeces^{-1}$) and $F$ is the daily diarrheal excretion rate (g faeces $person^{-1}$ $day^{-1}$).

To represent the summer population ($P$), months were selected where the minimum number of people on station was >30, and daily population values (n=601) were used as a discrete distribution, using data from 2005 to 2011. Daily per capita water use ($W$) was determined from monthly average population and monthly total station water use during summer months (2010-2011; AAD, 2012), with the variation represented by a Uniform distribution.

The secondary attack rate ($A_r$) is the proportion of people who, after contact with the original infected person, become ill (typically measured as the number of symptomatic cases). $A_r$ was used to estimate the maximum number of people who might be ill at one time (post-arrival of the one infected person), making the unrealistic (highly conservative) assumption that all infections occurred instantaneously (rather than over a period of days or weeks). Uniform distributions were used to represent the range of published values for secondary attack rate. Various studies have reported secondary norovirus attack rates between 0.14 and 0.22 over periods of up to 14 days after the first reported case (Alfano-Sobsey et al., 2012; Baron et al., 1982; Götz et al., 2002; Johansson et al., 2002; ter Waarbeek et al., 2010). Two studies reported very similar secondary attack rates for giardia

(Katz et al., 2006; Pickering et al., 1981) while a wide range (0 to 0.15) was reported for *Campylobacter* (Evans, 1996; Norkrans and Svedhem, 1982; Porter and Reid, 1980).

Shedding rates (*S*) were also represented by Uniform distributions. The only known study of giardia shedding rates ($S_G$; cysts person$^{-1}$ day$^{-1}$) was conducted with two infected individuals over a period of 7 weeks (Tsuchiya, 1931) and the maximum shedding rate from each participant was used to define the range of peak daily shedding rates. Lin et al. (2008) reported viable *Campylobacter* counts in faeces (CFU g$^{-1}$) from 10 samples while Faechem et al. (1983) reported counts as high as $10^9$ per g faeces (minimum and maximum values used to define the distribution). Three studies (Atmar et al., 2008; Chan et al., 2006; Lee et al., 2007) reported a range of norovirus shedding concentrations ($S_{NV}$; copies g-faeces$^{-1}$) and the maximum value from each of the four sets of data was used to define the distribution. For both norovirus and *Campylobacter*, a Uniform distribution (# g-faeces$^{-1}$) was converted to shedding rate using an estimate of daily diarrhoeal faecal weight (*F*; g person$^{-1}$ day$^{-1}$). Individuals suffering from diarrhea are typically defined as having a daily stool weight in excess of 200 g and a recent study reported mean stool weights of 750 g in persons with diarrhea (Rao, 2006); a Uniform distribution was used to represent faecal weights for ill individuals, making the assumption that all infected individuals have diarrhea (secondary attack rate counts only people who are symptomatic).

## 2.2 Population Size

The premise of this model is that small communities need to be considered differently to large cities, with the assumption that outbreak conditions will be significantly different to those in a large city as a function of the relatively greater degree of contact between community members in a small population and the greater level of dilution in a municipal sewage treatment plant due to the large population served (NRMMC et al., 2008). The estimate of municipal sewage concentrations reflects "average" conditions in a large city while outbreak sewage concentration was estimated assuming

228 that the community at Davis Station operates in a similar fashion to the confined populations
229 assessed to determine secondary attack rates (assuming a high degree of contact between all
230 community members). The difference in sewage concentrations during outbreaks in small or large
231 communities is a function of the proportion of the population infected. To evaluate the impact of
232 population size, a method was developed to estimate the likely sewage concentration, and therefore
233 required $\log_{10}$ reduction, during a norovirus outbreak in a large city. Norovirus was selected as the
234 reference pathogen as the required epidemiological data were available.
235
236 In Australia, there are 0.92 cases of gastroenteritis per person per year (Hall et al., 2006) of which
237 10.7% are caused by norovirus (Sinclair et al., 2005). If all norovirus infections occurred
238 simultaneously (which is highly improbable), then 9.8% of the population would be infected (~0.098
239 cases of novovirus infection per person per year). A more realistic scenario can be developed using
240 the results of a Melbourne study of 600 households that reported a maximum of 2.5% of households
241 with at least one case of norovirus per month (Sinclair pers. comm.; Sinclair et al., 2005), assuming
242 that monthly incidence rates equate to outbreaks. Assuming four people per household, 1.4 people
243 infected per household event (average value reported by Sinclair et al., 2005), and using the current
244 Melbourne population of 4,137,432 (ABS, 2012), an estimated 36,203 people would be infected
245 during an outbreak, or ~0.88% of the population. Applying this monthly infection rate across a whole
246 year, there would be 0.105 cases of norovirus per person per year which is consistent with the
247 estimated value above (0.098) and therefore a norovirus outbreak in Melbourne was conservatively
248 assumed to infect 1% of the population. The following scenarios were compared to evaluate the
249 magnitude of the effect of population size: municipal sewage (“average” city conditions), outbreak
250 conditions in a large city (population >1 million) and outbreak conditions at Davis Station.

## 2.3 Method Comparisons

The model presented herein uses a different approach to that taken by regulatory bodies. For example, a stochastic approach was used here to account for variability and uncertainty in the model while the Australian Guidelines for Water Recycling – Augmentation of Drinking Water Supplies (NRMMC et al., 2008) use a deterministic approach, conceding that stochastic analyses may provide a better understanding of uncertainty and variability where sufficient data is available. In our model, norovirus was chosen as the reference pathogen for viruses, giardia for protozoans and *Campylobacter* for bacteria, while the Guidelines use adenovirus measurements with the rotavirus dose-response model for viruses, cryptosporidium for protozoans and *Campylobacter* for bacteria. In addition, daily per capita drinking water consumption was much higher to reflect conditions at Davis Station. The differences in methods between the model used herein and that described in the Guidelines are outlined in Table S.2. The Guideline method and input parameters were used and then individual parameters were changed sequentially (detailed in Table 2, Table S.4 and Table S.5) to evaluate the impact of each change on the model output (required LRVs).

## 2.4 Sensitivity Analysis

A sensitivity analysis, using Spearman rank order correlation coefficients, was conducted using values from the first 1,000 random draws of each input distribution to identify those input parameters that had the greatest influence on the uncertainty of the model output. Input distributions were assessed to ensure there was no correlation between unrelated variables and then relevant input parameters were tested against the final model output (*LRV*). To further evaluate the impact of variation of input parameters on the magnitude of required *LRV*s, the model was run with key inputs set at discrete percentile values ($5^{th}$, $50^{th}$ and $95^{th}$), with no other alteration to the model; median required LRVs were reported.

## 2.5 Model Structure and Implementation

For all input parameters, a set of random values (n = 1,000,000) was drawn from the distribution and used for all model calculations. For all model outputs, the median and 90% confidence intervals

were reported. Confidence intervals were estimated using the percentile method (Buckland, 1984) and values are reported as follows: $50^{th}$ [$5^{th}$, $95^{th}$]; single values are $95^{th}$ percentile values unless otherwise indicated. Statistical differences were determined from the first 10,000 random draws from each output distribution using analysis of variance (ANOVA) and comparison of means using Tukey's HSD (Honestly Significant Difference) test. Differences were considered significant at $p \leq 0.05$. All modeling and analyses were performed in 'R' version 2.12.2 (The R Foundation for Statistical Computing, 2011) and some distribution fitting was conducted in @Risk (version 5.7).

# 3. Results

Estimates of norovirus, giardia and *Campylobacter* concentrations in municipal sewage ($1x10^7$, $9x10^3$ and $7.2x10^3$ # $L^{-1}$, respectively) were significantly lower ($p<0.0001$) than those determined for Davis Station outbreak conditions, $1.4x10^{12}$, $1.4x10^6$ and $4.9x10^8$ (Figure 1), which had a direct effect on the required LRVs. The required LRVs to meet the $\leq 10^{-6}$ DALYs $person^{-1}$ $year^{-1}$ health target, for potable reuse of treated wastewater at Davis Station, were 6.9 for norovirus, 8.0 for giardia and 7.4 for *Campylobacter* using estimates of municipal sewage, while for the Davis Station outbreak scenario they were 12.1, 10.4 and 12.3 respectively (Figure 2).

The estimate of norovirus concentration in municipal sewage ($1x10^7$ $L^{-1}$) was similar (within 1 order of magnitude) to many previously reported maximum sewage concentrations in Japan, UK, Italy, Finland, Germany, Sweden, Singapore and the Netherlands (Aw and Gin, 2010; Haramoto et al., 2006; Katayama et al., 2008; La Rosa et al., 2010; Laverick et al., 2004; Nordgren et al., 2009; Pusch et al., 2005; Van Den Berg et al., 2005; Von Bonsdorff et al., 2002), while the estimate of outbreak concentration ($1x10^{12}$ $L^{-1}$) was 5 orders of magnitude higher. Similarly, the estimate of giardia concentration in municipal sewage ($9x10^3$ $L^{-1}$) was within 1 order of magnitude of most of the previously reported maximum sewage concentrations in Japan, the Netherlands, Spain, Sweden and the USA (Castro-Hermida et al., 2008; Castro-Hermida et al., 2010; Gassmann and Schwartzbrod,

1991; Medema and Schijven, 2001; Oda et al., 2005; Ottoson et al., 2006a; Ottoson et al., 2006b; Sykora et al., 1991) while the estimate of giardia outbreak concentration ($1.4x10^{6}$ $L^{-1}$) was 3 orders of magnitude higher. The estimate of *Campylobacter* concentration in municipal sewage ($7.2x10^{3}$ cfu $L^{-1}$) was similar to published values from Italy and Spain (Rodríguez and Araujo, 2010; Stellacci et al., 2010), but lower (by as much as 2 orders of magnitude) than published concentrations in Germany and the Baltic Sea region (Holler, 1988; Rechenburg and Kistemann, 2009). The estimate of outbreak concentration ($4.9x10^{8}$) was up to 5 orders of magnitude higher than municipal sewage estimates.

The situation considered here is a worst case scenario where raw wastewater is not diluted with other wastewater sources (stormwater, rainwater, etc.). Each of the different scenarios and estimation methods had a significant effect ($p<0.001$) on the estimated sewage pathogen concentrations and subsequently the required LRVs. To evaluate the impact of population size on required LRVs, an epidemiological method was developed to estimate norovirus concentrations in Melbourne sewage during an outbreak. Melbourne outbreak sewage concentration ($7.2x10^{10}$ # $L^{-1}$) was nearly 4 orders of magnitude greater than municipal sewage ($1.0x10^{7}$ # $L^{-1}$) and ~1 order of magnitude less than Davis Station outbreak concentration ($1.4x10^{12}$ # $L^{-1}$), requiring 10.8 compared with 12.1 LRVs for Davis Station (Figure 3).

The Guidelines recommend a minimum enteric virus LRV of 9.5 for the production of drinking water from sewage while the model, using municipal sewage pathogen concentrations, determined a LRV of 6.9 for norovirus. To compare these two methods, sequential steps from the Guideline method to a deterministic approximation of the model are reported in Table 2. The difference in LRVs between Steps 2 and 4 shows that the full norovirus dose-response model reduces the required LRV from 12.5 with the rotavirus dose-response model to 7.2; this is likely the primary contributing factor to the difference between the Guideline value and the model value, although the higher virus

concentration was also important (increased the LRV from 9.4 to 12.5). The difference between Steps 4 and 5 shows the impact of using the higher drinking water volume (7.2 to 7.6) and the difference between Steps 5 and 7 shows the impact of a shorter exposure period (7.6 to 7.3); none of these changes greatly altered the final model output. Comparing the 95$^{th}$ percentile of the full stochastic model (6.9) with a deterministic approximation of the model (Step 7; 7.3), the difference is small, demonstrating that the understanding gained from the stepwise evaluation of parameter changes can be applied to the full model. A similar step-wise process was conducted for the other reference pathogens and results are presented in supplementary materials (Tables S.4 and S.5). The impact of the full stochastic model had much less impact on the LRVs for giardia and *Campylobacter*.

An assessment of all input parameters confirmed that there were no unexpected relationships or correlations and variation in many of the input parameters contributed significantly to the variation in the model outputs (Table 3). Using the municipal sewage method, sewage concentration had the largest impact on variation in the estimate of required LRVs, while drinking water volume, disease burden and exposure period contributed smaller amounts. Exposure period did not affect *Campylobacter*, while for giardia the dose-response parameter (*r*) and the infection to illness relationship also made significant contributions to variation. The outbreak scenario method was similar for norovirus and *Campylobacter*, with the greatest effect on variation in *LRV* due to variation in the estimate of sewage concentration which was a function of the other input parameters. Pathogen shedding rate contributed the most to the variation in LRVs for norovirus and *Campylobacter*, followed by faecal weight, disease burden, volume of drinking water and daily per capita water use. Secondary attack rate was also a significant contributor for *Campylobacter*. The variation in required LRVs for giardia was somewhat different and largely influenced by the variation in the dose-response parameter and illness to infection ratio, followed by drinking water volume, exposure period and daily per capita water use.

352 Table 2. Estimated required enteric virus $\log_{10}$ reduction values (LRVs) for stepwise methodological
353 changes from the Guideline method (NRMMC et al., 2008) to a deterministic approximation of the
354 model using municipal sewage concentrations.

| Step | LRV | Model Input Parameters[a] | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | *V* | *c* | *B* | $S_f$ | *inf:ill* | d-r | *n* |
| 1. | 9.4 | 2 | 8000 | $1.3x10^{-2}$ (RV) | 0.06 (RV) | 0.88 | RV[b] | 365 |
| 2. | 12.5 | 2 | $1.02x10^{7}$ ($95^{th}$ NV) | $1.3x10^{-2}$ (RV) | 0.06 (RV) | 0.88 | RV[b] | 365 |
| 3. | 9.8 | 4.8 ($95^{th}$ AAD) | 8000 | $1.3x10^{-2}$ (RV) | 0.06 (RV) | 0.88 | RV[b] | 365 |
| 4. | 7.2 | 2 | $1.02x10^{7}$ ($95^{th}$ NV) | $5.94x10^{3}$ ($95^{th}$ NV) | 0.99 ($95^{th}$ NV) | NV | NV[c] | 365 |
| 5. | 7.6 | 4.8 ($95^{th}$ AAD) | $1.02x10^{7}$ ($95^{th}$ NV) | $5.94x10^{3}$ ($95^{th}$ NV) | 0.99 ($95^{th}$ NV) | NV | NV[c] | 365 |
| 6. | 6.9 | 2 | $1.02x10^{7}$ ($95^{th}$ NV) | $5.94x10^{3}$ ($95^{th}$ NV) | 0.99 ($95^{th}$ NV) | NV | NV[c] | 118 ($95^{th}$ AAD) |
| 7. | 7.3 | 4.8 ($95^{th}$ AAD) | $1.02x10^{7}$ ($95^{th}$ NV) | $5.94x10^{3}$ ($95^{th}$ NV) | 0.99 ($95^{th}$ NV) | NV | NV[c] | 118 ($95^{th}$ AAD) |

355 [a]Model input parameters: *V* = daily water consumption (L person$^{-1}$), *c* = sewage pathogen
356 concentration (# L$^{-1}$), *B* = disease burden (DALYs case$^{-1}$), $S_f$ = susceptibility fraction, *inf:ill* = ratio of
357 infection to illness, d-r = dose-response model, *n* = days of exposure per year. $95^{th}$ refers to $95^{th}$
358 percentile of the input distribution. AAD = Davis Station data. NV = norovirus, RV = rotavirus.
359 [b]simplified approximate Beta-Poisson
360 [c]full Beta-Poisson
361

362 Table 3. Spearman's rank order correlation coefficients for required $\log_{10}$ pathogen reductions.

| Pathogen | Method | Model Input Parameters[a] | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | $c_{sewage}$ | $V$ | $S_f$ | $n$ | $B$ | $r$ | *inf:ill* | $P$ | $A_r$ | $S$ | $F$ | $W$ |
| Norovirus | municipal | 0.90[b] | 0.22[b] | -0.04 | 0.09[b] | 0.28[b] | n/a | n/a | n/a | n/a | n/a | n/a | n/a |
| | outbreak | 0.88[b,c] | 0.24[b] | -0.02 | 0.15[b] | 0.32[b] | n/a | n/a | -0.001 | 0.09[b] | 0.75[b] | 0.41[b] | -0.21[b] |
| Giardia | municipal | 0.86[b] | 0.20[b] | n/a[d] | 0.16[b] | 0.08[b] | 0.27[b] | 0.23[b] | n/a | n/a | n/a | n/a | n/a |
| | outbreak | 0.26[b,c] | 0.34[b] | n/a | 0.32[b] | 0.12[b] | 0.66[b] | 0.50[b] | -0.02 | 0.07[b] | 0.05 | n/a | -0.25[b] |
| *Campylobacter* | municipal | 0.99[b] | 0.07[b] | n/a | 0.00 | 0.14[b] | n/a | n/a | n/a | n/a | n/a | n/a | n/a |
| | outbreak | 0.93[b,c] | 0.23[b] | n/a | 0.09[b] | 0.25[b] | n/a | n/a | -0.07[b] | 0.45[b] | 0.66[b] | 0.33[b] | -0.16[b] |

363 [a]Model input parameters: $c_{sewage}$ = estimated sewage pathogen concentration (# $L^{-1}$), $V$ = daily water
364 consumption (L $person^{-1}$), $S_f$ = susceptibility fraction, $n$ = exposure period (days $year^{-1}$), $n$ = exposure
365 period (days $year^{-1}$), $B$ = disease burden (DALYs $case^{-1}$), $r$ = dose-response parameter for giardia,
366 *inf:ill* = ratio of infection to illness for giardia, $P$ = station population, $A_r$ = secondary attack rate, $S$ =
367 peak pathogen shedding, $F$ = daily faecal weight (g-faeces $person^{-1}$), $W$ = daily water use (L $person^{-1}$
368 $day^{-1}$).

369 [b]$p \leq 0.05$

370 [c]Outbreak sewage pathogen concentration was calculated from some or all of the following inputs:
371 station population, secondary attack rate, shedding rate, faecal weight, daily water use and dose-
372 response fit parameters. Its inclusion in the sensitivity analysis reflects the sum of variation
373 contributed by the other model input parameters.

374 [d]n/a = not applicable.

375

376 Similar trends were observed in the impact on LRVs when input parameters were fixed at discrete
377 percentile values (Figure 4). For municipal sewage scenarios, median LRVs were most affected by the
378 variation in the estimate of sewage concentration, with the spread in estimated LRVs as high as 2.3
379 $\log_{10}$ for giardia. For outbreak sewage scenarios, median LRVs were most affected by pathogen
380 shedding rate for norovirus and *Campylobacter* with a difference in LRVs as large as 1.3 $\log_{10}$
381 (*Campylobacter*). The effect of input parameter variation on LRVs for giardia was minimal for
382 outbreak conditions.

383 # 4. Discussion

384 While there have been recent arguments that the $10^{-6}$ DALY threshold is too conservative, even for
385 developed countries with lower background levels of water-borne disease (Mara, 2011; Mara et al.,
386 2010), the more cautious approach appears sensible in the context of small communities where, as a
387 result of isolation, the implications of illness may be much greater. Using the $10^{-6}$ DALY health target,
388 required LRVs were calculated to be 6.9, 8.0 and 7.4 for norovirus, giardia and *Campylobacter* using
389 municipal sewage values and 12.1, 10.4 and 12.3 for estimated Davis Station outbreak conditions,
390 compared with 9.5, 8.0 and 8.1 reported in the Guidelines (NRMMC et al., 2008). Using municipal
391 sewage concentrations, the LRVs for giardia and *Campylobacter* were very similar to the Guideline
392 values while the LRV for norovirus was much lower, largely due to the difference between the
393 rotavirus and norovirus dose-response models.

394

395 Under outbreak conditions, LRVs were much higher than Guideline values as a direct result of the
396 much higher sewage pathogen concentrations (3-5 orders of magnitude greater) estimated for Davis
397 Station outbreak conditions. These values, particularly norovirus, were orders of magnitude higher
398 than other published values of municipal sewage pathogen concentrations, reporting peaks of $10^3$—
399 $10^7$ for norovirus, $10^2$—$10^4$ for giardia and $10^4$—$10^7$ for *Campylobacter* (Table S.6). There is very little
400 information available on sewage pathogen concentrations during community gastroenteritis

401 outbreaks, although the Guidelines use 95$^{th}$ percentile values assumedly to represent peak pathogen
402 loads that might occur during an outbreak. To further evaluate the outbreak method, norovirus
403 concentrations at Davis Station were compared with estimated concentrations during an outbreak in
404 Melbourne. The proportion of people that become infected during a Melbourne norovirus outbreak
405 (1%) was much less than the secondary attack rate (14-18%) used for the Davis Station outbreak
406 scenario; therefore, Melbourne sewage was more dilute (i.e. lower pathogen concentration) and
407 required 10.8 compared with 12.1 LRVs for Davis. Assuming that the 95$^{th}$ percentile of the municipal
408 concentration estimate represents outbreak conditions, the median Melbourne outbreak
409 concentration ($2.6x10^{10}$ # $L^{-1}$) was nearly 3 orders of magnitude higher and may represent an
410 overestimation of outbreak concentrations. There are various possible explanations for this disparity
411 in concentration estimates: 1) the estimate of municipal sewage, based on data from Japan, does
412 not reflect Melbourne conditions (i.e. norovirus rates in Japan are lower than in Melbourne); 2) the
413 estimate of municipal sewage, based on monthly measurements, missed outbreak conditions; 3) the
414 outbreak method does not account for pathogen decay through the distribution system; or 4) the
415 outbreak sewage estimation method is too conservative. The impact of each of these potential
416 contributors cannot be quantified but importantly, even if the outbreak method overestimates
417 sewage concentration, the required LRVs are still higher than those in the Guidelines suggesting that
418 additional treatment will be required. A greater understanding of sewage pathogen concentrations
419 from small communities is needed to reduce the uncertainty around the estimated LRVs.
420
421 Various assumptions were made in the development of the model that may be important
422 constraints in the application of the model results. Secondary attack rate was used to estimate
423 outbreak sewage pathogen concentrations and is a measure of the spread of illness by direct
424 (person-to-person contact, inhalation of aerosols, etc.) and indirect (transfer from contaminated
425 surfaces, etc.) contact. Studies are typically conducted in relatively confined populations such as

426 households and school camps. While there is evidence that pathogen shedding can occur in the
427 absence of symptoms (Atmar et al., 2008; Birkhead and Vogt, 1989; Yakoob et al., 2010), the
428 secondary attack rate accounts for symptomatic cases only. Therefore, the model has not accounted
429 for asymptomatic infections that could contribute to the pathogen load in sewage. This may be of
430 limited concern, at least for norovirus, as recent investigations have found that asymptomatic cases
431 are unlikely to cause transmission despite high shedding rates (Sukhrie, 2012). We have also made
432 highly conservative assumptions that all individuals became ill instantaneously and shed pathogens
433 at the peak rate, and that all infected or ill individuals had diarrhea. In an actual outbreak, it is likely
434 that the spread of infection would occur over a few weeks (the time span of studies used to estimate
435 secondary attack rate). At the same time, pathogen shedding can occur for extended periods of time
436 – both prior to symptomatic illness and after apparent recovery – and it would seem unlikely that
437 peak shedding amongst all individuals would occur simultaneously.
438
439 Careful consideration will be required to design a treatment plant to meet safe drinking water
440 requirements in the event of an outbreak of gastroenteritis in a small community. The higher
441 required LRVs for norovirus, giardia and *Campylobacter* will demand a combination of treatment
442 systems. At Davis Station, a secondary treatment plant will be installed to remove the majority of
443 the wastewater contaminants, with additional tertiary and polishing treatment steps to meet
444 potable water quality requirements. The tertiary and polishing processes of large scale indirect
445 potable water systems generally consist of ultrafiltration, reverse osmosis and advanced oxidation
446 followed by final disinfection. Such systems provide a multi-barrier approach to ensure water quality
447 and are required to achieve a virus LRV of 9.5. Such processes can achieve higher LRVs (e.g. virus LRV
448 of 10 for Western Corridor in Brisbane, Australia), but nevertheless, the higher required LRVs for
449 small scale treatment plants as suggested by this model (e.g. an extra LRV of 2.6 for viruses) will

450 necessitate additional treatment units such as UV disinfection. The higher protozoa and bacteria
451 LRVs required for small systems also necessitate this extra treatment barrier.
452
453 In considering the higher required LRV requirements suggested by this model, it is important to
454 contextualize the risk of exposure to treated wastewater relative to other forms of exposure. A small
455 community such as Davis Station operates similar to a household in that the level of contact
456 between community members is quite high. The potential exposure pathways include person-to-
457 person contact, contact with contaminated surfaces and inhalation/ingestion of aerosols. The
458 assumption of the model, that one infected person arrives at Davis Station, would result in 18, 19 or
459 12 people sick with norovirus, giardia or *Campylobacter* respectively, based on the secondary attack
460 rate (direct or indirect contact with the infected person). In contrast, assuming all infected
461 individuals are shedding pathogens at a peak rate and that treatment of sewage conforms to the
462 required LRVs needed to meet the $10^{-6}$ DALY health target, consumption of the treated water would
463 result in up to 17 cases of norovirus, 5 cases of giardia or 2 cases of *Campylobacter* illness per 10,000
464 people or 0.18, 0.05 and 0.02 additional cases of norovirus, giardia and *Campylobacter* per summer
465 season (using $95^{th}$ percentile station population).
466
467 While Davis Station may be considered an extreme example, a similar approach could be applied to
468 many small remote communities in Australia. In the Northern Territory alone, there are 41
469 predominantly indigenous communities (95% indigenous) that range in size from 85 to 886
470 residents, with 13 of those communities having a population under 200 (ABS, 2007b). Other reports
471 have found that of the 1,139 remote indigenous communities across Australia, more than half (54%)
472 reported less than 20 residents and 23% reported populations of 20 to 49 (ABS, 2003). DPR may be
473 an appropriate solution in some of these communities and the results of this model demonstrate the
474 importance of consideration of small communities in determining appropriate treatment trains.

# 5. Conclusion

Direct potable reuse is a relatively new concept that has legitimate potential to enhance water security in both small and large communities. This analysis has highlighted the need to consider population size and vulnerability when assessing treatment requirements, a conclusion based on a quantitative microbial risk assessment (QMRA) that was conducted using norovirus, giardia and *Campylobacter* as reference pathogens. Two scenarios were compared, municipal sewage pathogen loads and potential pathogen loads during a community gastroenteritis outbreak, and pathogen concentrations were significantly higher ($p<0.001$) in the outbreak scenario. For the municipal sewage scenario, required LRVs were 6.9, 8.0 and 7.4 for norovirus, giardia and *Campylobacter* respectively, while for outbreak conditions, the values were 12.1, 10.4 and 12.3. While the outbreak values could overestimate LRVs by as much as 3 (for norovirus), they still indicate a need for additional treatment barriers for small communities in order to provide safe drinking water in the event of an outbreak. This higher treatment requirement is predominately attributed to the significantly increased pathogen levels in outbreak sewage relative to municipal sewage from a large city as a result of dilution and the relatively smaller proportion of the population infected. The recommended pathogen LRVs clearly represent a worst case scenario, assuming high pathogen concentrations and close community contact (high secondary attack rate). Generalization to other small communities is relevant nonetheless, and the model results indicate that in the event of an outbreak additional treatment barriers will be necessary to achieve safe drinking water in such communities.

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502

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